A FREEZE-DRY TECHNIQUE FOR THE PLANT COLLECTOR

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During a visit to the United States several years ago Dr. Hans-Christian Friedrich, Oberkonservator at the Munich Botanic Garden, described his work with the extremely succulent plants of Southwest Africa. In replying to a question on the preparation of herbarium specimens from such materials he reported on the success they had had in freezing the plants from a greenhouse collection in Munich and then drying the plant specimens in a plant press in the fashion familiar to collectors. The implication was made that the liquid from the plant tissues nearly "flowed out" of the defrosted specimens in the drying process. It was my feeling at that time that the technique might be useful also in meeting two distinct problems I had encountered in collecting plants in the American tropics. The first concerned the preparation of some of the fleshy or latex-possessing tropical plants encountered in field work in the Caribbean Islands. Specimens of the Cactaceae, Euphorbiaceae, Polygonaceae and Vitaceae usually dry slowly, fragment at nodes or articulations, discolor, become distorted in shape, or adhere to the drying papers. Suggestions have been made previously to overcome these problems involving treatment of specimens with solutions of formaldehyde, alcohol or chemical fixatives, or boiling in water. All of these techniques have been used at times with varying degrees of difficulty in the field and of success in the preparation of dried specimens.

The second problem occurs in the course of field work when the amount of rare or unusually high quality material to be pressed and dried exceeds the capacity of the drying apparatus. In tropical areas this material cannot be stored fresh in plastic bags or held loosely in buckets of water or even lightly contained in improvised presses without some deterioration of the specimens. In their Manual for Tropical Herbaria (Regnum Vegetabile 39: 45-48. 1965) Fosberg and Sachet summarize the various techniques that have been

developed in the tropics of both hemispheres for collecting plants in quantity and sending the material back to a distant base for arrangement later into individual specimens and for drying. These techniques call for arranging the specimens in papers alone or in presses and placing such bundles in plastic sheets, tubes or containers, or in metal cans to be sealed after alcohol, formaldehyde or other chemicals have been used to soak, paint, dust or immerse the specimens.

Both of these problems were encountered again during the summer of 1967 in the course of field work in St. Kitts and in Puerto Rico. Mr. Robert Wadsworth, who was collecting representative specimens on the former island as part of a study of the dwarfed elfin vegetation in the Antilles, worked with very limited drying facilities. At one stage of the operation a chest-type household freezer was made available through the courtesy of Mr. Colin Napier of St. Kitts. The plant specimens were prepared in the usual manner and the portion of the day's work which could not be accommodated on the small heater frame were placed in the freezer. Eventually, when space became available, the frozen press was removed from the freezer and placed directly on the heater. Now that all the specimens are mounted for insertion in the herbarium the frozen, then dried, specimens are indistinguishable in most cases from those dried directly from the field.

In Puerto Rico we had the opportunity of using the same technique while also making some comparative tests of the freezer-heater combination. Plants known to be difficult to prepare as specimens were deliberately chosen for trial. Mr. Joseph B. Martinson, who has supported many peculiar requests in relation to our study of the vegetation on Pico del Oeste, tolerated the use of his chest type of freezer. The cooperation of Dr. and Mrs. Richard J. Wagner and of Mr. Luis Maldonado helped complete the studies.

Our attempts to hold general collections for several days by freezing them and then completing the drying process were successful. There was some indication that the drying

process was slower than with freshly pressed specimens, but for the majority of collections the specimens prepared by the two methods were indistinguishable.

Specimens of certain native plants from the Luquillo Mountains and of certain species under cultivation by Mr. Martinson and at the Pennock Nurseries in Hato Rey were prepared in equal sets for comparative processing studies. One complete set was placed in a press and dried immediately, using electric space heaters with low-speed blowers. Other sets were prepared in presses and frozen for 48 hours. One of these was allowed to defrost to air temperature before it was placed on the heater, while a comparable set was placed on the heater directly from the freezer.

The fresh material which was placed directly over the heat without freezing dried from the lower side of the press, as expected, unless the specimens were rotated. The loss of liquid from the specimens was gradual and the separators rarely became very wet. The frozen material, however, tended to lose liquid quickly when defrosted so that the papers, blotters, or separators became saturated. Unless these were changed frequently the specimens often developed a growth of mold and the presses ultimately required 10-25% more time on the heater than did the fresh material. Recent correspondence with Dr. Friedrich reveals that he, too, experienced the problems of rot and fungus attack, which he controlled by soaking the press with alcohol after the first 24 hours of drying.

Only after several unsatisfactory attempts to use the freezer as an aid to plant collecting did we finally develop the technique and knowledge necessary to recommend it to those collectors fortunate enough to have a freezing compartment handy. For leathery or succulent specimens or those with thickened portions such as the large inflorescences of the Araceae or Musaceae, the stems or rhizomes of the Zingiberaceae, the rosette bases of the Bromeliaceae, or the pseudobulbs of the Orchidaceae, the freezing technique has real advantages. The specimens can be prepared whole or cut longitudinally and frozen either in newspaper

in the freezing compartment of a refrigerator, with dry ice in a chest or box, or in a loose press in a larger freezing unit. The frozen material is then returned to air temperature with the press loosely arranged. The liquid will often drain out of the specimens by gravity and the wet papers of the press must be changed. They now can be placed in a press and the pressure increased slightly or gradually for several hours to squeeze out more liquid before the specimens are finally placed on dry papers and arranged in a press for drying over heat. These specimens will dry more rapidly than fresh material since the liquid has been released from all of the tissues simultaneously. With this preliminary treatment the actual drying time is reduced 25 to 50%.

The benefits of this method of handling certain kinds of plant materials are evident on a comparative basis. There is a noticeable reduction in the fragmentation of stems at nodes, the disjunction of leaves from stems, the separation of parts of compound leaves, and the fragmentation of inflorescences. The parts of inflorescences of plants of *Heliconia* and *Musa* or various members of the Araceae or Zingiberaceae remain distinct, and single flowers or fruits can be lifted free when dry. This result contrasts dramatically with the agglutinated mass that is usually formed when the specimens are dried directly over heat. There is also less tendency for the specimens to adhere to the pressing papers. Succulent stems or woody ones which contain large amounts of liquid also dry more quickly after being frozen and tend to retain a more natural shape and color. There is much less distortion of fleshy parts of the plants if the pressure of the press is not excessive. Tracings were made of leaves of *Alloplectus* and of *Begonia* before and after drying and freeze-drying. Although both species showed the translucent characteristic of frozen material, the final leaf shape was less distorted and more truly representative of the fresh condition than was the material dried directly. The translucent condition of frozen-dried material even has an advantage in revealing more easily the vascular patterns.

In general, there was also a lessening of the darkening in those species which tend to darken on drying if the specimens had been frozen. There were, however, some notable exceptions.

The following materials have been treated by freeze-dry technique. Specimens cited bear the collecting numbers of the author and a representative set is deposited in the herbarium of the Arnold Arboretum, Harvard University.

Alocasia sp. Araceae *(17041)*. A stout herb under cultivation with very thick inflorescence and rhizome. The stems, inflorescence, and petiole sections retained a more characteristic shape after freezing.

Alloplectus ambiguus Urb. Gesneriaceae *(16641, 16815)*. An epiphytic herb with succulent stems and foliage. The frozen specimens have a translucent appearance, while heat-dried specimens tend to darken. The fleshy fruit is much less distorted after freezing.

Anthurium dominicense Schott. Araceae *(16642)*. An epiphytic rosette-forming plant with succulent and leathery leaves. Heat-dried specimens turn a pale brown color, while freezing causes a blackening of the leaves. The rhizomes retained more natural shape after freezing. All specimens proved to be very susceptible to fungus attack during drying.

Begonia decandra Par. Begoniaceae *(16939)*. A terrestrial herb with normally pink-colored translucent succulent stems and leaves. Heat-dried specimens tended to darken, while those frozen before heat drying became more translucent and dried a lighter color. Fragmentation of the leaves from the stems or of the inflorescense was conspicuously less in the material which had been frozen.

Caryota sp. Palmae *(16912)*. A cultivated stout cane-type fishtail palm. Both inflorescences and leaves were prepared. The frozen leaves retained a green color while the fresh material darkened. The flowers and young fruits are retained on the rachi much more securely in the frozen material. The mature flowers, however, fell from the rachus in both methods of preparation.

Cissus trifoliata L. Vitaceae. An herbaceous vine which has succulent leaves and stems. The fresh dried specimens showed the fragmentation botanists have come to expect with most tropical species of *Cissus*. A considerable reduction in fragmentation was evident in the specimens which had been frozen. The leaflets were retained intact in most cases and the stem fragmented only at the very young nodes.

Clusia grisebachiana (Pl. & Tr.) Alain. Guttiferae *(16632)*. Small tree with thick coriaceous leaves and copious latex in tissues of leaf and stem cortex. Fruits thick and inflorescence commonly fragile. Specimens dried noticeably faster after freezing and retained a clearer yellow-tan color. The fragility was not reduced in any noticeable manner.

Epidendrum ramosum Jacq. Orchidaceae *(16640)*. Epiphytic. This species of orchids tends to break apart in normal drying. The frozen specimens retained their leaves in more satisfactory numbers, but the specimens were abnormally translucent when dried.

Forsteronia portoricensis Woodson. Asclepiadaceae *(16637)*. This is a rampant vine with heavy semi-fleshy leaves, abundant latex and deep carmine red flowers. The fresh-dried specimens showed the dark brown or black discoloration often obtained with plants having a latex. The specimens which had been frozen first became an unexpected greenish-brown color.

Guzmannia berteroniana (R. & S.) Mez Bromeliaceae *(16638)*. An epiphytic rosette plant with conspicuously bracted inflorescence. Following the usual practice the rosette was reduced to one quarter the original size before processing. In some specimens the inflorescence was split down the middle before drying. This plant does not produce beautiful herbarium specimens with any technique. The fresh-dried material showed the straw-brown color characteristic of most herbarium specimens. The frozen material retained a greenish cast. The colorful bracts and flowers discolored in both processes but the parts were more easily separated in the frozen specimens.

Hedyosmum arborescens Sw. Chloranthaceae *(16633)*. A tree with fragile branches, abundant liquid in the stems, succulent leaves, and white fleshy perianth parts to the fruit. All Caribbean material of *Hedyosmum* shrinks noticeably in drying and turns a characteristic chocolate brown. In this test the frozen material showed a slight translucence not noticeable in heater-dried material.

Heliconia bihai L. Musaceae *(16643)*. A clump-forming herb with succulent petioles and a thick distichous inflorescence. Material was prepared of the leaf blade and the petiole base, and of the inflorescence, which was split longitudinally. *Heliconia* specimens are notoriously difficult to dry. Several collectors have attempted to boil the specimens before drying, with very unsatisfactory specimens resulting. In this test all frozen material dried much more rapidly and made much better looking specimens. The frozen leaves retained a better color. The frozen inflorescense was flatter, but the individual flowers could be separated easily and the fruits remained distinct, very much in contrast to the heater-dried material.

Hillia parasitica Jacq. Rubiaceae *(16634)*. An epiphytic or climbing succulent woody plant with pendant branches. The long white tubular flowers are fleshy and the leaves are thick. The specimens dried by heat alone showed the characteristic black color of most herbarium specimens, while the flowers are an ivory white color. Following freezing, however, the leafy specimens were less discolored and a lighter green color suggesting a translucence, while all of the flowers were either pale tan or brown with a definite translucent aspect.

Musa sapientum L. Musaceae *(16938)*. A succulent herb with massive inflorescence. Herbarium specimens of banana inflorescences are infrequent in herbaria. Very young inflorescences were split lengthwise for this test, while older ones were separated into bract and flower components for both pistillate and staminate flowers. The heater-dried material discolored and became distorted in shape. The frozen material dried more quickly, showed similar discoloration but much less distortion.

Philodendron aff. *bipinnatifidum* Araceae *(16928)*. A heading type of *Philodendron*. Inflorescences were split and frozen, then defrosted and placed in press for drying. Regular drying methods produced much-distorted specimens in which the flowers and fruits were tightly agglutinated. Freezing produced equally darkened specimens, slightly less distorted but with separate flowers and young fruits. Leaves dried and darkened in both techniques.

Philodendron krebsii Schott. Araceae *(16644)*. A high-climbing herbaceous vine with succulent stems and leaves and a fleshy inflorescence. Normal heater drying produced the usual blackened specimens and agglutinated inflorescences. Material which had been frozen before drying was tan or light brown in color and the spathe and spadix separated more readily for dissection.

Pitcairnia angustifolia L. Bromeliaceae *(16636)*. A rosette-forming terrestrial plant with stout inflorescence axil and red succulent fruits. Heater-dried material turned a straw-brown color, while the frozen and dried material retained the natural glaucous-gray color. In flower and fruit, however, the plant parts showed better natural pigmentation when dried directly.

Plumeria alba L. Apocynaceae. The stout stems of *Plumeria* contain copious amounts of a low viscosity latex which causes specimens dried normally to adhere to paper. The flowers are fleshy and in normal drying techniques discolor slightly and usually fall free from the inflorescence. The Puerto Rican specimens tend to have the leaves more tightly curled or involute than those of other West Indian populations. The frozen dried material produced noticeably flatter leaves with a slight degree of translucence. The frozen flowers were conspicuously translucent but were not held on the inflorescence to any greater degree. Frozen material generally did not adhere to the drying papers.

Vriesia sintenisii (Baker) Sm. & Pittend. Bromeliaceae *(16639)*. An epiphytic plant with a rosette of leaves and a stout erect rhizome. This species under normal preparation does not make attractive specimens. Those dried from

fresh condition showed considerable shriveling of the leaves, contraction of the inflorescence, and general darkening of the specimen. The frozen material, in contrast, retained a more natural form of rosette and leaves, and all parts showed more natural pigmentation.

Not every expedition to a tropical area will be fortunate enough to have a freezer with space available at a base camp. When such circumstances do occur or when it is necessary to prepare unusually succulent materials in other surroundings, the freezer may aid the collector in holding massed collections and in speeding the drying while improving the technique for processing difficult materials.

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ARNOLD ARBORETUM
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